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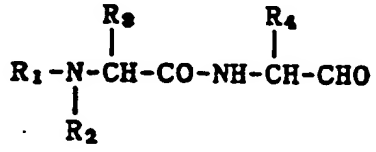
(54) Proteinase inhibitor.

(57) A proteinase inhibitory compound represented by the following general formula (1):



(1)

wherein R_1 represents a straight-chain or branched acyl group having 2 to 10 carbon atoms, a branched-, cyclic- or polycyclic-alkyloxycarbonyl group having 4 to 15 carbon atoms, a substituted- or unsubstituted-benzyloxycarbonyl group, a 2,2,2-trichloroethyloxycarbonyl group, a 2-(trimethylsilyl)ethyloxycarbonyl group, a p-toluenesulfonyl group, an o-nitrophenylsulfenyl group, a diphenylphosphonothioyl group, a triphenylmethyl group or a 2-benzoly-1-methylvinyl group;
R_2 represents a hydrogen atom; or R_1 and R_2 may together form a phthaloyl group;
R_3 represents an isobutyl group, a n-butyl group or an isopropyl group and the above-mentioned R_1 can b an unsubstituted-benzyloxycarbonyl group provided that R_3 is a n-butyl group; and
R_4 represents a n-butyl group.

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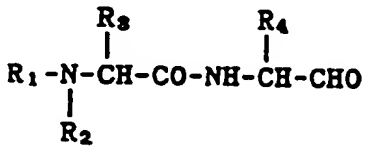
PROTEINASE INHIBITOR

This invention relates to a novel N-substituted peptidyl aldehyde r presented by the general formula (1) which will be given hereinafter. Furthermore, it relates to a cysteine proteinase inhibitor which compris s a compound of the general formula (1) as an active ingredient and exerts an intense enzyme inhibitory activity on cysteine proteinases, in particular, on papain, calpain and even on cathepsin.

It has been desired to develop a drug which specifically inhibits the activities of papain (E.C.3.4.22.2) and calpain (E.C.3.4.22.17) which are cysteine proteinases, since such a drug is useful as an anti-inflammatory agent. Calpain, which widely occurs in mammals and birds, is observed in cytosol. It is considered that abnormal activation of this enzyme causes serious diseases including muscular dystrophy and cataracts. Cathepsin (E.C.3.4.22.1) is a proteinase localized in lysosome. It is considered that abnormal activation of this enzyme causes cancer metastasis, amyotrophy and muscular dystrophy. Therefore, it has been desired to develop a drug which specifically inhibits the activities of calpain and cathepsin and thus is applicable as a remedy for muscular dystrophy, amyotrophy and cataracts or as a metastasis inhibitor. Attempts have been made to develop drugs useful for the above-mentioned purposes. As a result, a number of cysteine proteinase inhibitors have been found out hitherto [Shimizu B. et al., J. Antibiot., 25, 515 (1972); Japanese Patent Laid-Open No. 28990/1985; Japanese Patent Laid-Open No. 106600/1986; and Japanese Patent Laid-Open No. 103897/1986]. However, each of the known inhibitors required to be further improved in respect of activity and biomigration. Furthermore, it is urgently required to develop a compound which also exerts an intense inhibition activity on cathepsin.

The present invention provides a compound which exerts a strong inhibition activity on cyst ine proteinases, in particular, cathepsin B and which shows excellent biomigration.

More particularly, the present invention provides a novel N-substituted peptidyl aldehyde represented by the following general formula (1):



(1)

wherein R_1 represents a straight-chain or branched acyl group having 2 to 10 carbon atoms (e.g., octanoyl, caproyl or isovaleryl group), a branched, cyclic or polycyclic alkyloxycarbonyl group having 4 to 15 carbon atoms (e.g., t-butyloxycarbonyl, adamantyloxycarbonyl or isobornyloxycarbonyl group), a benzyloxycarbonyl group optionally substituted with, e.g., a halogen atom, a nitro group or a methoxy group, a 2,2,2-trichloroethyloxycarbonyl group, a 2-(trimethylsilyl)ethyloxycarbonyl group, a p-toluenesulfonyl group, an o-nitrophenylsulfenyl group, a diphenylphosphonothioyl group, a triphenylmethyl group or a 2-benzoyl-1-methylvinyl group;

R_2 represents a hydrogen atom; or R_1 and R_2 may together form a phthaloyl group;

R_3 represents an isobutyl group, a n-butyl group or an isopropyl group and the above-mentioned R_1 can be an unsubstituted-benzyloxycarbonyl group provided that R_3 is a n-butyl group; and

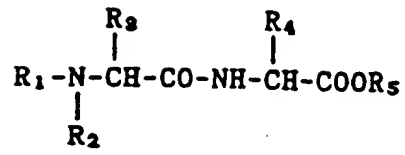
R_4 represents a n-butyl group;

which exerts a strong activity of inhibiting cathepsin B, calpain or papain.

The peptide derivative compound of the present invention represented by the general formula (1) strongly inhibits papain, calpain and cathepsin B. Thus, it is expected that this compound will further inhibit other cysteine proteinases, for example, cathepsin H or L. Furthermore, it is expected that this compound would be useful in the treatment of inflammation, cataracts, epidermolysis bullosa and pemphigus which might be caused by these cysteine proteinases. Furthermore, an affinity column prepared by coupling the compound of the present invention on an appropriate support is available in the purification of cysteine proteinases. In addition, it is expected that the compound of the present invention would be available as a reagent in the fields of biochemistry and enzymology.

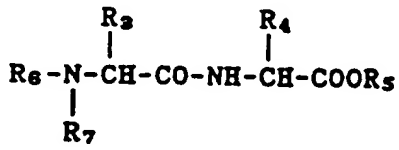
The compound of the present invention may be prepared in the following manner.

First, a compound of the general formula (1) wherein a group represented by R_1 or a group formed by R_1 together with R_2 is stable in treatment with a base or reduction with a reducing agent may be prepared as follows. A compound represented by the following general formula (2):



(2)

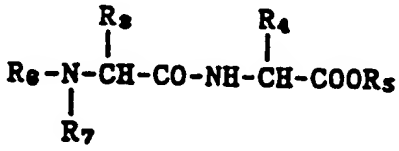
wherein R_1, R_2, R_3 and R_4 are as defined above regarding the general formula (1); and
R_5 represents a lower alkyl group;
is reduced by use of a reducing agent in an organic solvent so as to give an alcohol compound. Next, th obtained alcohol compound is oxidized by use of an oxidizing agent to thereby give the aldehyde. Alternatively, a compound represented by the following general formula (3):



(3)

wherein R_3, R_4 and R_5 are as defined above regarding the above general formula (2);
R_6 represents an amino-protecting group which is stable to a treatment with a base or reduction by use of a reducing agent; and
R_7 represents a hydrogen atom; or
R_6 and R_7 together form an amino-protecting group which is stable to a treatment with a base or reduction by use of a reducing agent;
can be used as the starting compound. The protecting group represented by R_6 or formed by R_6 together with R_7 is removed by an appropriate method. A desired group R_1 or a desired group formed by R_1 together with R_2 is then introduced to the amino group to give a compound represented by the general formula (2), which is then converted to a compound of the general formula (1) as described above.

A compound represented by the general formula (1) wherein a group represented by R_1 or a group formed by R_1 together with R_2 is unstable into a treatment with a base or reduction by use of a reducing agent may be prepared as follows. A compound represented by the following general formula (3):



(3)

wherein R_3, R_4 and R_5 are as defined above regarding the above general formula (2);
R_6 represents an amino-protecting group which is stable to a treatment with a base or reduction by use of a reducing agent; and
R_7 represents a hydrogen atom; or
R_6 and R_7 together form an amino-protecting group which is stable to a treatment with a base or reduction by use of a reducing agent;
is reduced by use of a reducing agent in an organic solvent so as to give an alcohol compound. Next, the protecting group R_6 or the one formed by R_6 together with R_7 is removed by an appropriate method. Then a desired R_1 group or the one formed by R_1 together with R_2 is introduced into the amino group by an appropriate method. The alcohol moiety of the compound thus obtained is then oxidized in the sam manner as the one described above so as to give the aldehyde. Thus the aimed compound can be readily obtained.

Examples

To further illustrate the present invention, the following enzyme inhibition activity tests and Examples will be given. It is needless to say, h wever, that the technical scope of th present invention is not restricted thereby.

Example 1

Production of N-octanoyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $CH_3(CH_2)_6CO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-octanoyl-L-leucine

2.6 g of L-leucine was dissolved in 20 ml of 1 N sodium hydroxide. 3.3 ml of octanoyl chloride and 20 ml of 1 N sodium hydroxide were added thereto under ice-cooling and the mixture so formed was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with ether. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The extract was dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus, 4.3 g of the titled N-octanoyl-L-leucine was obtained in the form of crystals.

(b) Production of N-octanoyl-L-leucyl-L-norleucine methyl ester

2.6 g of the N-octanoyl-L-leucine obtained in the above step (a) and 1.8 g of L-norleucine methyl ester hydrochloride were dissolved in 50 ml of dry dimethylformamide. Next, 1.6 g of diethyl cyanophosphonat was added thereto. 2.0 g of triethylamine was further added to the obtained solution under ice-cooling and the resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, 200 ml of water was added to the reaction mixture. Then it was extracted with ether. The organic phase was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.2 g of the titled N-octanoyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-octanoyl-L-leucyl-L-norleucinol

2.2 g of the N-octanoyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 20 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 8 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 1.7 g of the titled N-octanoyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-octanoyl-L-leucyl-L-norleucinal

1.7 g of the N-octanoyl-L-leucyl-L-norleucinol obtained in the above step (c) and 2.0 g of triethylamine were dissolved in 20 ml of dry dimethylsulfoxide. Then a solution of 3.0 g of sulfur trioxide/pyridine complex dissolved in 20 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room

temperature for 10 minutes and th n poured into 200 ml of ice water. Aft r thric extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, th residue was recrystallized from a solvent mixtur of ethyl acetate and hexane. Thus 1.0 g of th target compound N-octanoyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 2

Production of N-caproyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $CH_3(CH_2)_4CO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-caproyl-L-leucine

2.6 g of L-leucine was dissolved in 20 ml of 1 N sodium hydroxide. 3.0 g of caproyl chloride and 20 ml of 1 N sodium hydroxide were added thereto under ice-cooling and the mixture so formed was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with ether. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The extract was dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was recrystallized from a solvent mixtur f ethyl acetate and hexane. Thus, 2.8 g of the titled N-caproyl-L-leucine was obtained in the form of crystals.

(b) Production of N-caproyl-L-leucyl-L-norleucine methyl ester

2.7 g of the N-caproyl-L-leucine obtained in the above step (a) and 2.2 g of L-norleucine methyl ester hydrochloride were dissolved in 50 ml of dry dimethyformamide. Next, 2.0 g of diethyl cyanophosphonate was added thereto. 2.0 g of triethylamine was further added to the obtained solution under ice-cooling and the resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, 200 ml of water was added to the reaction mixture. Then it was extracted with ether. The organic phase was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.2 g of the titled N-caproyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-caproyl-L-leucyl-L-norleucinol

2.2 g of the N-caproyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 20 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 80 °C. Next, 8 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 1.6 g of the titled N-caproyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-caproyl-L-leucyl-L-norleucinal

1.6 g of the N-caproyl-L-leucyl-L-norleucinol obtained in the above step (c) and 2.0 g of triethylamine were dissolved in 15 ml of dry dimethylsulfoxide. Then a solution of 3.0 g of sulfur trioxide/pyridine complex

dissolved in 15 ml of dimethylsulfoxid was added thereto under stirring. Th mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate under reduced pressure, th residue was purified by medium pressure column chromatography by use of silica gel. Thus 1.0 g of the target compound N-caproyl-L-leucyl-L-norleucinal was obtained in the form of powder.

Example 3

Production of N-isovaleryl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $(CH_3)_2CHCH_2CO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-isovaleryl-L-leucine

5.2 g of L-leucine was dissolved in 40 ml of 1 N sodium hydroxide. 2.4 g of isovaleryl chloride and 40 ml of 1 N sodium hydroxide were added thereto under ice-cooling and the mixture so formed was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with either. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or bel w followed by extracting with ethyl acetate. The extract was dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 2.6 g of the titled N-isovaleryl-L-leucine was obtained in the form of crystals.

(b) Production of N-isovaleryl-L-leucyl-L-norleucine methyl ester

2.6 g of the N-isovaleryl-L-leucine obtained in the above step (a) and 2.2 g of L-norleucine methyl ester hydrochloride were dissolved in 50 ml of dry dimethylformamide. Next, 2.0 g of diethyl cyanophosphonat was added thereto. 2.0 g of triethylamine was further added to the obtained solution under ice-cooling and the resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, 200 ml of water was added to the reaction mixture. Then it was extracted with ether. The organic phase was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 1.8 g of the titled N-isovaleryl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-isovaleryl-L-leucyl-L-norleucinol

1.8 g of the N-isovaleryl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 30 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 5 ml of absolute methanol was added thereto dropwise under reflux. After th completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 1.2 g of the titled N-isovaleryl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-isovaleryl-L-leucyl-L-norleucinal

1.0 g of th N-isovaleryl-L-leucyl-L-norleucinol obtained in the above step (c) and 2.0 g of triethylamine w re dissolved in 15 ml of dry dim thylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridin complex dissolved in 15 ml of dimethylsulfoxide was added thereto und r stirring. The mixture was stirred at room temperature for 10 minutes and th n poured into 200 ml of ice wat r. Aft r thrice extracting with thyl acetate, it was successively washed with a 10% aqueous soluti n of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.7 g of the target compound N-isovaleryl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 4

Production of N-t-butyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $(CH_3)_3COCO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-t-butyloxycarbonyl-L-leucyl-L-norleucine methyl ester

2.5 g of N-t-butyloxycarbonyl-L-leucine monohydrate was dissolved in 50 ml of dry methylene chloride. Then 1.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To th solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 6 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 4.0 g of the titled N-t-butyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(b) Production of N-t-butyloxycarbonyl-L-leucyl-L-norleucinol

3.0 g of the N-t-butyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) and 3.0 g of sodium borohydride were suspended in 60 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 10 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-t-butyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(c) Production of N-t-butyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-t-butyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (b) and 1.2 g of triethylamine were dissolved in 12 ml of day dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide was added thereto under stirring. Th mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.5 g of the target compound N-t-butyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 5

Production of N-adamantyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1). R_1 = tricyclo-[3.3.1.13.7]decane-OCO-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-adamantyloxycarbonyl-L-leucine

2.6 g of L-leucine and 7.0 g of potassium carbonate were dissolved in 100 ml of water. 4.0 g of adamantyloxycarbonyl fluoride was added thereto under ice-cooling and the mixture so formed was stirred at room temperature for 3 hours. After the completion of the reaction, the reaction mixture was washed with ether. 50% phosphoric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ether. The extract was dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 3.8 g of the titled N-adamantyloxycarbonyl-L-leucine was obtained in the form of crystals.

(b) Production of N-adamantyloxycarbonyl-L-leucyl-L-norleucine methyl ester

3.1 g of the N-adamantyloxycarbonyl-L-leucine obtained in the above step (a) was dissolved in 50 ml of dry methylene chloride. 1.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was add d thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride. The mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.8 g of the titled N-adamantyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-adamantyloxycarbonyl-L-leucyl-L-norleucinol

2.6 g of the N-adamantyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90° C. Next, 10 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.2 g of the titled N-adamantyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of a powder.

(d) Production of N-adamantyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-adamantyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.0 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. Then a solution of 1.6 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was purified by reverse phase medium pressure column chromatography by use of octadecyl silane. Thus 0.8 g of the target compound N-adamantyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of a powder.

Example 6

Production of N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = (4-Cl)-$C_6H_4CH_2OCO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-p-chlorobenzyloxycarbonyl-L-leucine dicyclohexylamine salt

3.9 g of L-leucine and 4.2 g of potassium carbonate were dissolved in 100 ml of water. A solution of 6.2 g of p-chlorobenzyloxycarbonyl chloride dissolved in 10 ml of dioxane was added thereto under ice-cooling. The mixture so formed was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with ether. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The extract was dried over anhydrous sodium sulfate. After concentrating the solvent under reduced pressure, 6.0 ml of dicyclohexylamine was added thereto. Thus 9.5 g of the titled N-p-chlorobenzyloxycarbonyl-L-leucine, dicyclohexylamine salt was obtained in the form of crystals.

(b) Production of N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester

4.8 g of the N-p-chlorobenzyloxycarbonyl-L-leucine dicyclohexylamine salt obtained in the above step (a) was suspended in 100 ml of ethyl acetate and washed with a 10% aqueous solution of citric acid. Th organic phase was dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was dissolved in 50 ml of methylene chloride. 2.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution thus formed, was added a solution of 1.8 g of L-norleucine methyl ester and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride followed by stirring at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 3.0 g of the titled N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(c) Production of N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol

3.0 g of the N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 40 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90° C. Next, 10 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetat , washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressur column chromatography by use of silica gel. Thus 2.2 g of the titled N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of an oily product.

(d) Production of N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.2 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 10 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off th ethyl acetate, the

residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.7 g of th target compound N-p-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 7

Production of N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = (4-CH_3O)-$C_6H_4CH_2OCO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-p-methoxybenzyloxycarbonyl-L-leucine dicyclohexylamine salt

3.9 g of L-leucine and 7.0 g of triethylamine were dissolved in 15 ml of water. A solution of 9.1 g of p-methoxybenzyl-S-4, 6-dimethylpyrimidin-2-ylthiocarbonate dissolved in 20 ml of dioxane was added thereto under ice-cooling. The mixture so formed was stirred at room temperature for 5 hours. After the completion of the reaction, 150 ml of water was added to the reaction mixture followed by washing with ethyl acetate. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The extract was dried over sodium sulfate. After distilling off the solvent under reduced pressure, 6.0 ml of dicyclohexylamine was added to the concentrated solution. Thus 9.0 g of the titled N-p-methoxybenzyloxycarbonyl-L-leucine dicyclohexylamine salt was obtained in the form of crystals.

(b) Production of N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester

4.7 g of the N-p-methoxybenzyloxycarbonyl-L-leucine dicyclohexylamine salt obtained in the above step (a) was suspended in 100 ml of ethyl acetate and washed with a 10% aqueous solution of citric acid. Th organic phase was dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was dissolved in 50 ml of dry methylene chloride. 2.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimid hydrochloride was added thereto. To the solution thus formed, was added a solution of 1.8 g of L-norleucin methyl ester hydrochloride and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride followed by stirring at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 3.0 g of the titled N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(c) Production of N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinol

2.0 g of the N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 40 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 10 ml of absolute methanol was added thereto dropwis under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the abov step (c) and 1.2 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur

trioxide/pyridine complex dissolved in 10 ml of dimethylsulfoxid was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ic water. After thrice extracting with ethyl acetate, it was successiv ly washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrog ncarbonat and a saturated aqueous solution of NaCl and dried over anhydrous solution sulfate. After distilling off th thyl acetate, th residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.7 g of the target compound N-p-methoxybenzyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 8

Production of N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = (4-NO_2)-$C_6H_4CH_2OCO$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester

7.4 g of N-benzyloxycarbonyl-L-leucine was dissolved in 100 ml of dry methylene chloride and 8.2 g f 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 7.4 g of L-norleucine methyl ester hydrochloride and 4.2 g of triethylamine in 100 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After th completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 12 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(b) Production of N-benzyloxycarbonyl-L-leucyl-L-norleucinol

10 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) and 3.0 g of sodium borohydride were suspended in 100 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90 °C. Next, 20 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 100 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 8.0 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(c) Production of N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinol

2.5 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (b) was dissolved in 50 ml of ethanol. Then a catalytic amount of palladium carbon was added thereto and the mixture so formed was stirred under a hydrogen atmosphere for 3 hours. After the completion of the reaction, th palladium carbon was filtered off and the solvent was distilled off from the filtrate under reduced pressur . Thus L-leucyl-L-norleucinol was quantitatively obtained. Next, 20 ml of water and 1.5 g of sodium hydrogencarbonate were added thereto. Further, a solution of 1.5 g of p-nitrobenzyloxycarbonyl chloride dissolved in 10 ml of ether was added thereto followed by stirring at room temperature for 3 hours. After the completion of the reaction, the mixture was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and then dried over anhydrous magnesium sulfate. After distilling off the ethyl acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 2.0 g of the titled N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the abov step (c) and 1.2 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 10 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.8 g of the target compound N-p-nitrobenzyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 9

Production of N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = (2-Cl)-$C_6H_4CH_2OCO$-, R_2 = H-, R_3 = $(CH_3)_2OHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-o-chlorobenzyloxycarbonyl-L-leucine dicyclohexylamine salt

4.0 g of L-leucine ethyl ester hydrochloride and 2.1 g of triethylamine were dissolved in 100 ml of dry tetrahydrofuran. Then 5.0 g of N-(2-chlorobenzyloxycarbonyl)oxysuccinimide was added thereto under ice-cooling. The mixture so formed was stirred at room temperature for 12 hours. After the completion of the reaction, the solvent was distilled off under reduced pressure and 50 ml of water was added to the residue. After extracting with ethyl acetate, the extract was dried over sodium sulfate. The solvent was distilled off under reduced pressure and the residue was purified with medium pressure column chromatography by use of silica gel. Thus 7.0 g of N-o-chlorobenzyloxycarbonyl-L-leucine ethyl ester was obtained in the form of an oily product. Next, this product was dissolved in 20 ml of methanol and 30 ml of 1 N sodium hydroxide and 50 ml of water were added thereto. The mixture so formed was heated to 80°C under stirring for 1 hour. After the completion of the reaction, the reaction mixture was washed with ethyl acetate. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The organic phase was dried over anhydrous sodium sulfate and concentrated under reduced pressure. After adding 6.0 g of dicyclohexylamine, 5.5 g of the titled N-o-chlorobenzyloxycarbonyl-L-leucine dicyclohexylamine salt was obtained in the form of crystals.

(b) Production of N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester

4.8 g of the N-o-chlorobenzyloxycarbonyl-L-leucine dicyclohexylamine salt obtained in the above step (a) was suspended in 100 ml of ethyl acetate and washed with a 10% aqueous solution of citric acid. The organic phase was dried over sodium sulfate. After distilling off the solvent, the residue was dissolved in 50 ml of methylene chloride. 2.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution thus formed, was added a solution of 1.8 g of L-norleucine methyl est r hydrochloride and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride followed by stirring at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 3.0 g of the titled N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(c) Production of N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol

3.0 g of the N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.0 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated under

reflux in a nitrogen atmospher at 90°C. Next, 10 ml of absolute m thanol was added thereto dropwise und r reflux. After the compl tion of the addition, the mixture was stirred under reflux for 30 minut s and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl ac tate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magn sium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal

1.0 g of the N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.2 g of triethylamine were dissolved in 15 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 15 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, th residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.8 g of the target compound N-o-chlorobenzyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 10

Production of N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = CCl_3CH_2OCO-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-benzyloxycarbonyl-L-leucyl-norleucine methyl ester

7.4 g of N-benzyloxycarbonyl-L-leucine was dissolved in 100 ml of dry methylene chloride and 8.2 g of 1-ethyl-3-(3-diethyl-aminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 7.4 g of L-norleucine methyl ester and 4.2 g of triethylamine dissolved in 100 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 12 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(b) Production of N-benzyloxycarbonyl-L-leucyl-L-norleucinol

10 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) and 3.0 g of sodium borohydride were suspended in 100 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 20 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 100 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 8.0 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(c) Production of N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinol

2.5 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (b) was dissolved in 50 ml of ethanol. Then a catalytic amount of palladium carbon was added thereto and the mixture so formed was stirred under a hydrogen atmosphere for 3 hours. After th compl tion of the reaction, the palladium carbon was filtered off and the solvent was distilled off from the filtrate und r reduced pressure. Thus L-leucyl-L-norleucinol was quantitatively obtained. Next, 20 ml of water and 2.2 g of sodium hydrogencarbonate were added thereto. Further, a solution of 1.5 g of 2,2,2-trichloroethyloxycarbonyl chloride in 10 ml of ether was added thereto followed by stirring at room temperature for 3 hours. After the completion of the reaction, the mixture was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and then dried over anhydrous magnesium sulfate. After distilling off the ethyl acetate, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 1.0 g of the titled N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinal

0.7 g of the N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.0 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. Then a solution of 1.5 g of sulfur trioxide/pyridine complex dissolved in 10 ml of dimethylsulfoxide was added thereto under stirring. Th mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.5 g of the target compound N-(2,2,2-trichloroethyl)oxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 11

Production of N-(trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $(CH_3)_3SiCH_2CH_2$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-OCO-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of p-nitrophenyl trimethylsilylethyloxycarbonate

4.4 g of trimethylsilylethanol and 10 g of bis(p-nitrophenyl) carbonate were dissolved in 100 ml of dry methylene chloride and 6.0 g of N-methylmorpholine was added thereto. The mixture so formed was stirr d at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 0.1% sulfuric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 5.0 g of the titled p-nitrophenyl trimethylsilylethyloxycarbonate was obtained in the form of crystals.

(b) Production of N-trimethylsilylethyloxycarbonyl-L-leucine dicyclohexylamine salt

4.0 g of L-leucine methyl ester hydrochloride and 2.2 g of triethylamine were dissolved in 100 ml of dry dimethylformamide and 5.8 g of p-nitrophenyl trimethylsilylethyloxycarbonate was added thereto. To the solution so formed, was added 200 mg of 1-hydroxybenzotriazole monohydride as a catalyst. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was concentrated under reduced pressure and water was added. After extracting with ethyl acetat , the organic phase was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 7.0 g of the N-trimethylsilylethyloxycarbonyl-L-leucine methyl ester was obtained in the form of crystals. The obtained

product was dissolved in 20 ml of methanol and 30 ml of a 1 N sodium hydroxide and 50 ml of water were added thereto. The mixture was stirred at 80° C for 1 hour. After the completion of th reaction, the mixture was washed with ethyl acetate and 0.1% of sulfuric acid was added to th aqueous phase to thereby lower the pH valu to 2 or below. After extracting with ethyl acetate, th organic phase was dri d over anhydrous sodium sulfate and concentrated under reduced pressure. After adding 6.0 g of dicyclohexylamine, 5.5 g of the titled N-trimethylsilylethyloxycarbonyl-L-leucine dicyclohexylamine salt was obtained in the form of crystals.

(c) Production of N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucine methyl ester

4.6 g of the N-trimethylsilylethyloxycarbonyl-L-leucine dicyclohexylamine salt obtained in the above step (b) was suspended in 100 ml of ethyl acetate. After washing with a 10% aqueous solution of citric acid, the organic phase was dried over anhydrous sodium sulfate. The solvent was distilled off under reduced pressure and the N-trimethylsilylethyloxycarbonyl-L-leucine thus obtained was dissolved in 50 ml of dry methylene chloride. Next, 2.0 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.2 g of triethylamine in 50 ml of dry methylene chloride. The mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 10% citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.8 g of the titled N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(d) Production of N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinol

2.8 g of the N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucine methyl ester and 1.5 g of sodium borohydride were suspended in 40 ml of t-butyl alcohol. The resulting mixture was heated under reflux at 90° C under a nitrogen atmosphere. Then 12 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the reaction mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated solution of NaCl and dried over anhydrous magnesium sulfate. After distilling off the ethyl acetate, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(e) Production of N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinal

1.8 g of the N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (d) and 1.9 g of triethylamine were dissolved in 20 ml of dry dimethylsulfoxide. To the solution so formed, was added a solution of 3.0 g of sulfur trioxide/pyridine complex dissolved in 20 ml of dimethylsulfoxide. After stirring at room temperature for 20 minutes, the mixture was poured into 300 ml of ice water, thrice extracted with ethyl acetate, successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the obtained residue was purified by reverse phase medium pressure column chromatography by use of octadecyl silane. Thus 1.0 g of the target compound N-trimethylsilylethyloxycarbonyl-L-leucyl-L-norleucinal was obtained in the form of an oily product.

Example 12

Production of N-p-toluenesulfonyl-L-leucyl-L-norleucinal [In general formula (1), R_1 = $(4\text{-}CH_3)C_6H_4SO_2$-, R_2

= H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-p-toluenesulfonyl-L-leucine

2.6 ml of L-leucine was dissolved in 40 ml of 2 N sodium hydroxide. 3.8 g of p-toluenesulfonyl chloride was added thereto under ice-cooling and the mixture so formed was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with ethyl acetate. 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below followed by extracting with ethyl acetate. The organic phase was dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 3.6 g of the titled N-p-toluenesulfonyl-L-leucine was obtained in the form of crystals.

(b) Production of N-p-toluenesulfonyl-L-leucyl-L-norleucine methyl ester

1.8 g of the N-p-toluenesulfonyl-L-leucine obtained in the above step (a) was dissolved in 50 ml of dry methylene chloride and 1.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.2 g of triethylamine dissolved in 50 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressur column chromatography by use of silica gel. Thus 2.8 g of the titled N-p-toluenesulfonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-p-toluenesulfonyl-L-leucyl-L-norleucinol

2.8 g of the N-p-toluenesulfonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.5 g of sodium borohydride were suspended in 40 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90 °C. Next, 12 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 30 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-p-toluenesulfonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-p-toluenesulfonyl-L-leucyl-L-norleucinal

1.0 g of the N-p-toluenesulfonyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.2 g of triethylamine were dissolved in 12 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide was added thereto under stirring. Th mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, th residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.7 g of the target compound N-p-toluenesulfonyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 13

Production of N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinal. [in general formula (1), R_1 = $(2\text{-}NO_2)C_6H_4S$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-o-nitrophenylsulfenyl-L-l ucin dicyclohexylamine salt

5.0 g of L-leucine and 8.0 g of o-nitrophenylsulfenyl chloride were dissolved in 25 ml of a 2 N solution of sodium hydroxide and 25 ml of 1,4-dioxane was added thereto. The mixture so formed was stirred at room temperature for 1 hour. After the completion of the reaction, 2 N sulfuric acid was added to the reaction mixture followed by extracting with ether. The organic phase was dried over sodium sulfate. After distilling of the solvent under reduced pressure, 8.0 g of dicyclohexylamine was added. Thus 10 g of the titled N-o-nitrophenylsulfenyl-L-leucine dicyclohexylamine salt was obtained in the form of crystals.

(b) Production of N-o-nitrophenylsulfenyl-L-leucyl-L-norleucine methyl ester

4.6 g of the N-o-nitrophenylsulfenyl-L-leucine dicyclohexylamine salt obtained in the above step (a) was suspended in 100 ml of ethyl acetate and washed with a 10% aqueous solution of citric acid. Then the organic phase was dried over sodium sulfate. After distilling off the solvent, the obtained N-o-nitrophenylsulfenyl-L-leucine was dissolved in 50 ml of dry methylene chloride and 2.0 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.0 g of triethylamine dissolved in 50 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with a 10% citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 5.0 g of the titled N-o-nitrophenylsulfenyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinol

3.2 g of the N-o-nitrophenylsulfenyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 2.0 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90 °C. Next, 15 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 50 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinal

1.0 g of the N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.2 g of triethylamine were dissolved in 12 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide was added thereto under stirring. Th mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was purified by reverse phase medium pressure column chromatography by use of octadecyl silane. Thus 1.2 g of the target compound N-o-nitrophenylsulfenyl-L-leucyl-L-norleucinal was obtained in the form of an oily product.

Example 14

Production of N-diphenylphosphonothioyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $(C_6H_5)_2PS$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-diphenylphosphonothioyl-L-leucyl-L-norleucine methyl ester

2.0 g of N-diphenylphosphonothioyl-L-leucine was dissolved in 50 ml of dry methylene chloride and 2.0 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ester hydrochloride and 1.0 g of triethylamine dissolved in 50 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 10% citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 3.0 g of the titled N-diphenylphosphonothioyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(b) Production of N-diphenylphosphonothioyl-L-leucyl-L-norleucinol

2.5 g of the N-diphenylphosphonothioyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) and 1.0 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90° C. Next, 10 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 50 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetat , washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-diphenylphosphonothioyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(c) Production of N-diphenylphosphonothioyl-L-leucyl-L-norleucinal

1.0 g of of the N-diphenylphosphonothioyl-L-leucyl-L-norleucinol obtained in the above step (b) and 0.8 g of triethylamine were dissolved in 8 ml of dry dimethylsulfoxide. To the solution so formed, was added a solution of 1.3 g of sulfur trioxide/pyridine complex in 8 ml of dimethylsulfoxide. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice-water. After thrice extracting with ethyl acetate, the extract was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residu was purified by reverse phase medium pressure column chromatography by use of octadecyl silane. Thus, 0.6 g of the target compound N-diphenylphosphonothioyl-L-leucyl-L-norleucinal was obtained in the form f an oily product.

Example 15

Production of N-triphenylmethyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $(C_6H_5)_3C$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-benzyloxycarbonyl-L-leucyl-norleucine methyl ester

7.4 g of N-benzyloxycarbonyl-L-leucine was dissolved in 100 ml of dry methyl n chloride and 8.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimid hydrochloride was added thereto. T th solution so formed, was added a solution of 7.4 g of L-norleucine methyl ester hydrochloride and 4.2 g of triethylamine dissolved in 100 ml of dry methylene chloride. The resulting mixture was stirred at room temp rature for 12 hours. After the completion f the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonat and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 12 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(b) Production of N-triphenylmethyl-L-leucyl-L-norleucine methyl ester

4.0 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) was dissolved in 50 ml of ethanol. Then a catalytic amount of palladium carbon was added thereto and th mixture so formed was stirred under a hydrogen atmosphere for 3 hours. After the completion of th reaction, the palladium carbon was filtered off and the solvent was distilled off from the filtrate under reduced pressure. Thus L-leucyl-L-norleucine methyl ester was quantitatively obtained. Next, this product was dissolved in 100 ml of dry methylene chloride and 1.5 g of triethylamine was added thereto. Further, 4.2 g of triphenylmethane chloride was added and the resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 4.3 g of the tiled N-triphenylmethyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(c) Production of N-triphenylmethyl-L-leucyl-L-norleucinol

4.3 g of the N-triphenylmethyl-L-leucyl-L-norleucine methyl ester obtained in the above step (b) and 1.5 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90°C. Next, 15 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 100 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.8 g of the titled N-triphenylmethyl-L-leucyl-L-norleucinol was obtained in the form of powdery crystals.

(d) Production of N-triphenylmethyl-L-leucyl-L-norleucinal

1.0 g of the N-triphenylmethyl-L-leucyl-L-norleucinol obtained in the above step (c) and 2.1 g of triethylamine were dissolved in 10 ml of dry dimethylsulfoxide. To the solution so formed, was added a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 10 ml of dimethylsulfoxide. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice-water. After thrice extracting with ethyl acetate, the extract was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate.

After distilling off the ethyl acetate, the obtained residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 0.7 g of the target compound N-triphenylmethyl-L-leucyl-L-norleucinal was obtained in the form of crystals.

Example 16

Production of N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinal [in general formula (1), R_1 = $C_6H_5COCH=C(CH_3)$-, R_2 = H-, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-(2-benzoyl-1-methyl)vinyl-L-leucine

6.6 g of benzoylacetone was dissolved in 100 ml of dry ethanol. To the solution so formed, was added a solution of 1.8 g of sodium hydroxide dissolved in 20 ml of dry methanol. 5.4 g of L-leucine was furth r added thereto and the resulting mixture was heated under reflux at 100° C for 3 hours. After the completion of the reaction, the solvent was concentrated under reduced pressure and water was added. The pH value of the reaction mixture was lowered to around 2 with 10% citric acid. Then the mixture was extracted with ether and dried over anhydrous sodium sulfate followed by distilling off the solvent under reduced pressure. The residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 7.0 g of the titled N-(2-benzoyl-1-methyl)vinyl-L-leucine was obtained in the form of crystals.

(b) Production of N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucine methyl ester

2.8 g of the N-(2-benzoyl-1-methyl)vinyl-L-leucine obtained in the above step (a) was dissolved in 50 ml of dry methylene chloride and 2.0 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 1.8 g of L-norleucine methyl ster hydrochloride and 1.0 g of triethylamine dissolved in 50 ml of dry methylene chloride. The mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 10% citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and then dried over sodium sulfat . After distilling off the solvent under reduced pressure, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 3.5 g of the titled N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucine methyl ester was obtained in the form of an oily product.

(c) Production of N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinol

2.5 g of the N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucine methyl ester obtained in the above st p (b) and 1.0 g of sodium borohydride were suspended in 50 ml of t-butyl alcohol and then heated und r reflux in a nitrogen atmosphere at 90° C. Next, 10 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 50 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.0 g of the titled N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinal

1.0 g of the N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinol obtained in the above step (c) and 1.2 g of triethylamine were dissolved in 12 ml of dry dimethylsulfoxide. Then a solution of 2.0 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide was added thereto under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, it was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl acetate, the residue was purified by reverse phase medium pressure column chromatography by use of octadecyl silane. Thus 0.8 g of the target compound N-(2-benzoyl-1-methyl)vinyl-L-leucyl-L-norleucinal was obtained in th form of an oily product.

Example 17

Production of N-phthaloyl-L-leucyl-L-norleucinal [in general formula (1), R_1 and R_2 form a 1,2-$(CO-)_2C_6H_4$ group together, R_3 = $(CH_3)_2CHCH_2$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester

7.4 g of N-benzyloxycarbonyl-L-leucine was dissolved in 100 ml of dry methylene chloride and 8.2 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 7.4 g of L-norleucine methyl ester hydrochloride and 4.2 g of triethylamine dissolved in 100 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 12 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester was obtained in the form of crystals.

(b) Production of N-benzyloxycarbonyl-L-leucyl-L-norleucinol

10 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucine methyl ester obtained in the above step (a) and 3.0 g of sodium borohydride were suspended in 100 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90° C. Next, 20 ml of absolute methanol was added thereto dropwise under reflux. After the completion of the addition, the mixture was stirred under reflux for minutes and then cooled to room temperature. 100 ml of water was added thereto under ice-cooling. After distilling off the methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 8.0 g of the titled N-benzyloxycarbonyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(c) Production of N-phthaloyl-L-leucyl-L-norleucinol

2.5 g of the N-benzyloxycarbonyl-L-leucyl-L-norleucinol obtained in the above step (b) was dissolved in 50 ml of ethanol and a catalytic amount of palladium carbon was added thereto. The mixture was stirred under a hydrogen atmosphere for 3 hours. After the completion of the reaction, the palladium carbon was filtered off and the solvent was distilled off from the filtrate. Thus L-leucyl-L-norleucinol was quantitatively obtained. To the product so formed, were added 20 ml of water, 2.2 g of potassium carbonate and 1.6 g of carboethoxyphthalimide. The resulting mixture was stirred at room temperature for 2 hours. After the completion of the reaction, the mixture was thrice extracted with ethyl acetate, washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. After distilling off the ethyl acetate, the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 0.5 g of the titled N-phthaloyl-L-leucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-phthaloyl-L-leucyl-L-norleucinal

0.5 g of the N-phthaloyl-L-leucyl-L-norleucinol obtained in the above step (c) and 0.7 g of triethylamine were dissolved in 6 ml of dry dimethylsulfoxide. To the solution so formed, was added a solution of 1.0 g of sulfur trioxide/pyridine complex dissolved in 6 ml of dimethylsulfoxide under stirring. The resulting mixture was stirred at room temperature for 10 minutes and poured into 100 ml of ice water. Then it was thrice extracted with ethyl acetate, ssuccessively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off th solvent, the residue was recrystallized from a solvent mixture of thyl acetate and hexane. Thus 0.3 g of the target compound N-

phthaloyl-L-leucyl-L-norleucinal was obtained in th form of crystals.

Example 18

Production of N-benzyloxycarbonyl-L-norleucyl-Lnorleucinal [in general formula (1), R_1 = $C_6H_5CH_2OCO$-, R_2 = H-, R_3 = $CH_3(CH_2)_3$-, R_4 = $CH_3(CH_2)_3$-]:

(a) Production of N-benzyloxycarbonyl-L-norleucine

7.8 g of L-norleucine and 8.4 g of potassium carbonate were dissolved in 200 ml of water. Then a solution of 12.4 g of benzyloxycarbonyl chloride dissolved in 20 ml of dioxane was added thereto under ice-cooling. The resulting mixture was stirred at room temperature for 8 hours. After the completion of the reaction, the reaction mixture was washed with ether and 5 N hydrochloric acid was added to the aqueous phase to thereby lower the pH value to 2 or below. After extracting with ethyl acetate, the organic phase was dried over anhydrous sodium sulfate and the solvent was distilled off. Thus 9.5 g of the titled N-benzyloxycarbonyl-L-norleucine was obtained in the form of an oily product.

(b) Production of N-benzyloxycarbonyl-L-norleucyl-L-norleucine methyl ester

7.0 g of N-benzyloxycarbonyl-L-norleucine obtained in the above step (a) was dissolved in 100 ml of dry methylene chloride and 5.4 g of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride was added thereto. To the solution so formed, was added a solution of 4.2 g of L-norleucine methyl ester hydrochloride and 5.2 g of triethylamine in 100 ml of dry methylene chloride. The resulting mixture was stirred at room temperature for 12 hours. After the completion of the reaction, the reaction mixture was successively washed with 1 N hydrochloric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the solvent, the residue was purified by medium pressure column chromatography by use of silica gel. Thus 8.0 g of the titled N-benzyloxycarbonyl-L-norleucyl-L-norleucine methyl ester was obtained in the form of crystals.

(c) Production of N-benzyloxycarbonyl-L-norleucyl-L-norleucinol

4.0 g of the N-benzyloxycarbonyl-L-norleucyl-L-norleucine methyl ester obtained in the above step (b) and 1.2 g of sodium borohydride were suspended in 100 ml of t-butyl alcohol and then heated under reflux in a nitrogen atmosphere at 90° C. Next, 18 ml of absolute methanol was added thereto dropwise und r reflux. After the completion of the addition, the mixture was stirred under reflux for 30 minutes and then cooled to room temperature. 50 ml of water was added thereto under ice-cooling. After distilling off th methanol and t-butyl alcohol under reduced pressure, the residue was thrice extracted with ethyl acetat , washed with a saturated aqueous solution of NaCl and dried over anhydrous magnesium sulfate. The ethyl acetate was distilled off under reduced pressure and the obtained residue was purified by medium pressure column chromatography by use of silica gel. Thus 2.2 g of the titled N-benzyloxycarbonyl-L-norleucyl-L-norleucinol was obtained in the form of crystals.

(d) Production of N-benzyloxycarbonyl-L-norleucyl-L-norleucinal

1.7 g of the N-benzyloxycarbonyl-L-norleucyl-L-norleucinol obtained in the above step (c) and 1.8 g of triethylamine were dissolved in 12 ml of dry dimethylsulfoxide. To the solution so formed, was added a solution of 3.0 g of sulfur trioxide/pyridine complex dissolved in 12 ml of dimethylsulfoxide under stirring. The mixture was stirred at room temperature for 10 minutes and then poured into 200 ml of ice water. After thrice extracting with ethyl acetate, the extract was successively washed with a 10% aqueous solution of citric acid, a saturated aqueous solution of NaCl, a saturated solution of sodium hydrogencarbonate and a saturated aqueous solution of NaCl and dried over anhydrous sodium sulfate. After distilling off the ethyl

acetate, the residue was recrystallized from a solvent mixture of ethyl acetate and hexane. Thus 1.2 g of the target compound N-benzyloxycarbonyl-L-norleucyl-L-norleucinal was obtained in the form of crystals.

Table 1 shows the physical properties of each compound.

Table 1

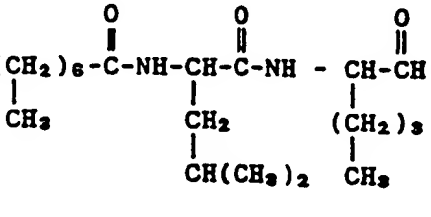
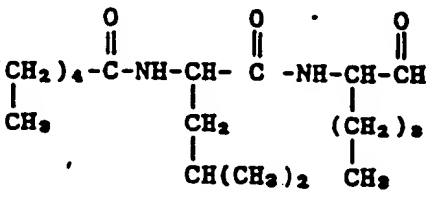
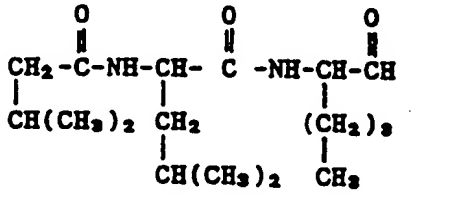
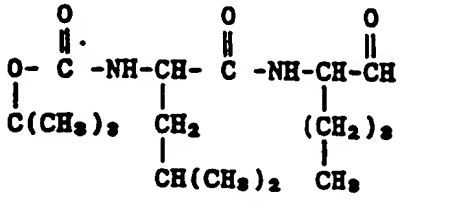
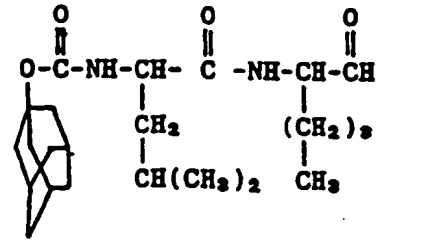
Example No.	Structure	Physical Property (^{1}H-NMR: $CDCl_3$, standard TMS, ppm)
1		0.80-1.20(12H,m), 1.20-2.10(19H,m), 2.20(2H,t,J=8.0Hz), 4.20-4.70(2H,m), 6.08(1H,d,J=8.0Hz), 6.92(1H,d,J=7.0Hz), 9.54(1H,s) m.p. 55°C (decompose)
2		0.80-1.10(12H,m), 1.10-2.00(15H,m), 2.18(2H,t,J=8.0Hz), 4.26(1H,dd,J=8.0Hz, J=12Hz), 4.66(1H,dd, J=8.0Hz,J=14Hz), 6.92(1H,d,J=8.0Hz), 7.82(1H,d,J=8.0Hz), 9.50(1H,s)
3		0.80-1.20(15H,m), 1.20-2.00(10H,m), 2.00-2.30(2H,m), 4.20-4.80(2H,m), 6.12(1H,d,J=8.0Hz), 7.00(1H,d,J=7.0Hz), 9.50(1H,s) m.p. 134°C (decompose)
4		0.80-1.05(9H,m), 1.20-2.00(9H,m), 1.46(9H,s),4.12(1H, dd,J=8.0Hz,J=15Hz), 4.48(1H,dd,J=8.0Hz, J=13Hz), 4.84(1H,d, J=8.0Hz), 6.64(1H,d, J=7.0Hz), 9.56(1H,s) m.p. 57°C (decompose)
5		0.80-1.10(9H,m), 1.20-2.20(9H,m), 1.65(6H, broad-s), 2.12(9H, broad-s), 4.14(1H,dd, J=6.0Hz,J=14Hz), 4.48 (1H,dd,J=7.0Hz,J=13Hz) 4.84(1H,d,J=6.0Hz), 5.64(1H,d,J=7.0Hz), 9.56(1H,s)

Table 1 (cont'd)

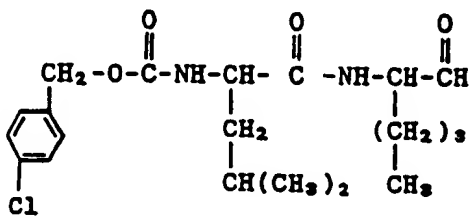
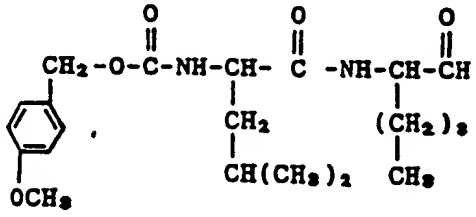
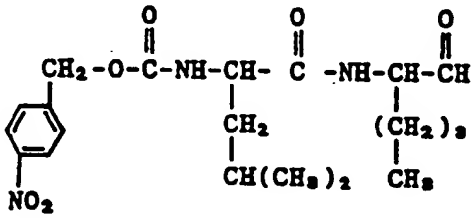
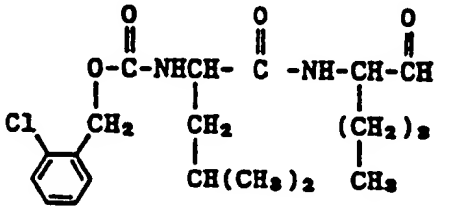
Example No.	Structure	Physical Property (^{1}H-NMR: $CDCl_3$, standard TMS, ppm)
6		0.80-1.11(9H,m), 1.10-2.00(9H,m), 4.20(1H, dd,J=8.0Hz,J=12Hz), 4.48(1H,dd,J=8.0Hz, J=11Hz), 5.06(2H,s), 5.14(1H,d,J=8.0Hz), 6.44(1H,d,J=8.0Hz), 7.30(4H,s), 9.55(1H,s) m.p. 93°C (decompose)
7		0.80-1.00(9H,m), 1.10-2.00(9H,m), 3.82(3H,s), 4.20(1H,m) 4.48(1H,dd,J=7.0Hz, J=12Hz), 5.04(2H,s), 5.08(1H,d,J=7.0Hz), 6.48(1H,d,J=7.0Hz), 6.86(2H,d,J=9.0Hz), 7.28(2H,d,J=9.0Hz), 9.54(1H,s) m.p. 87°C (decompose)
8		0.80-1.10(9H,m), 1.10-2.00(9H,m), 4.20(1H, dd,J=8.0Hz,J=14Hz), 4.52(1H,dd,J=6.0Hz, J=11Hz), 5.20(2H,s), 5.32(1H,d,J=8.0Hz), 6.46(1H,d,J=6.0Hz), 7.48(2H,d,J=8.0Hz), 8.20(2H,d,J=8.0Hz), 9.56(1H,s) m.p. 89°C (decompose)
9		0.70-1.10(9H,m), 1.10-2.00(9H,m), 4.20(1H,m), 4.48(1H, dd,J=6.0Hz,J=13Hz), 5.18(1H,d,J=6.0Hz), 5.24(2H,s), 6.48(1H, d,J=8.0Hz), 7.10-7.50 (4H,m), 9.56(1H,s) m.p. 114°C (decompose)

Table 1 (cont'd)

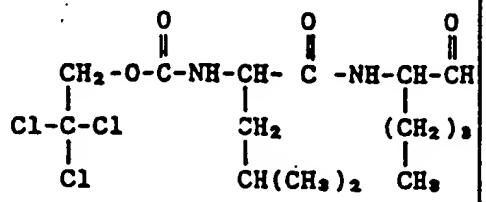
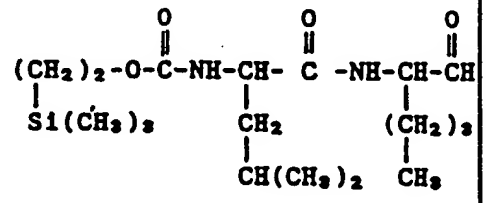
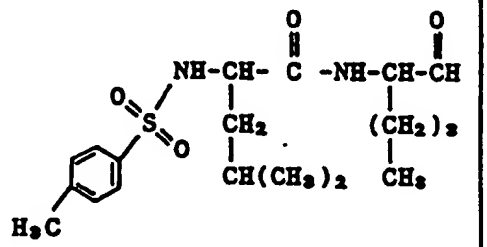
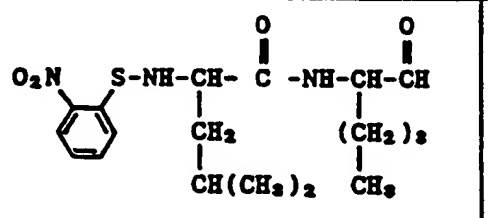
Example No.	Structure	Physical Property (^{1}H-NMR: $CDCl_3$, standard TMS, ppm)
10		0.80-1.00(9H,m), 1.10-2.00(9H,m), 4.20(1H, dd,J=8.0Hz,J=16Hz), 4.53(1H,dd,J=7.0Hz, J=11Hz), 4.74(2H,s), 5.42(1H,d,J=8.0Hz), 8.40(1H,d,J=7.0Hz), 9.58(1H,s) m.p. 86°C (decompose)
11		0.00(9H,s), 0.80-1.10 (12H,m), 1.10-2.00 (9H,m), 4.12(2H,dd, J=8.0Hz,J=10Hz), 4.18(1H,m), 4.44(1H, dd, J=7.0Hz,J=13Hz), 5.13(1H,d,J=7.0Hz), 8.68(1H,d,J=8.0Hz), 9.52(1H,s) Refractive index = 1.4634
12		0.73(3H,d,J=6.0Hz), 0.86(3H,d,J=6.0Hz), 0.90(3H,t,J=6.0Hz), 1.10-1.90(9H,m), 2.42 (3H,s), 3.74(1H,dd, J=8.0Hz,J=13Hz), 4.30 (1H,dd,J=7.0Hz,J=14Hz) 5.10(1H,d,J=8.0Hz), 8.46(1H,d,J=7.0Hz), 7.28(2H,d,J=8.0Hz), 7.74(2H,d,J=8.0Hz), 9.54(1H,s) m.p. 135°C (decompose)
13		0.70-1.10(9H,m), 1.10-2.20(9H,m), 4.00(1H,m), 4.56(1H, dd,J=7.0Hz,J=13Hz), 4.70(1H,m), 6.56(1H,d, J=7.0Hz), 7.26(1H,m), 7.62(1H,m), 8.02(1H,m) 8.24(1H,m), 9.56(1H,s)

Table 1 (cont'd)

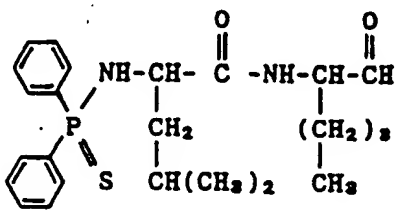
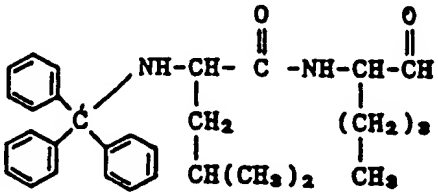
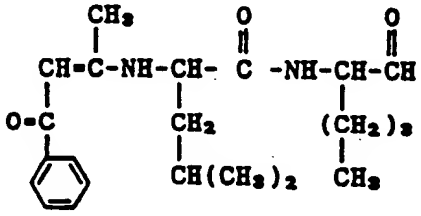
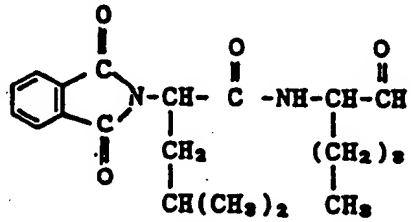
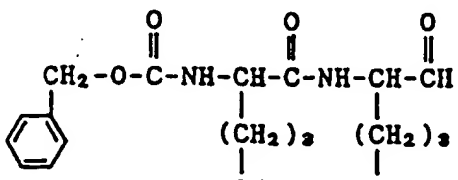
Example No.	Structure	Physical Property (^{1}H-NMR: $CDCl_3$, standard TMS, ppm)
14		0.70-1.00(9H,m), 1.00-2.00(9H,m), 3.40(1H, dd,J=7.0Hz,J=9.0Hz), 3.92(1H,m), 4.28(1H, dd,JNH-CH=7.0Hz, JPNH=13Hz), 6.64(1H, d,J=7.0Hz), 7.30-7.60 (6H,m), 7.70-8.00 (4H,m), 9.46(1H,s) Refractive index = 1.5692
15		0.70-1.00(9H,m), 1.10-2.00(9H,m), 3.36(1H,m), 4.08(1H,m), 7.10-7.50(15H,m), 9.20(1H,s) m.p. 123°C (decompose)
16		0.70-1.10(9H,m), 1.10-2.00(9H,m), 2.12(3H,s) 4.12(1H,dd,J=8.0Hz, J=16Hz), 4.52(1H,dd, J=8.0Hz,J=13Hz), 5.86(1H,s), 6.60(1H,d, J=8.0Hz), 7.20-7.60 (3H,m), 7.70-8.00 (2H,m), 9.54(1H,s), 11.38(1H,d,J=8.0Hz) Refractive index = 1.5814
17		0.80-1.10(9H,m), 1.10-2.00(8H,m), 2.40(1H,m), 4.54(1H, dd,J=7.0Hz,14Hz), 5.00(1H,dd,J=5.0Hz, J=11Hz), 6.86(1H,d, J=7.0Hz), 7.70-8.00 (4H,m), 9.58(1H,s) m.p. 96°C (decompose)

Table 1 (cont'd)

Example No.	Structure	Physical Property (^{1}H-NMR: $CDCl_3$, standard TMS, ppm)
18		0.80-1.00(6H,m), 1.10-2.00(12H,m), 4.20(1H, dd,J=8.0Hz,J=14Hz), 4.52(1H,dd,J=7.0Hz, J=13Hz), 5.12(2H,s), 5.32(1H,d,J=8.0Hz), 6.56(1H,d,J=7.0Hz), 7.38(5H,s), 9.56(1H,s) m.p. 128°C (decompose)

Example 19

Enzyme Inhibitory Activity of the Invention Compound

The enzyme inhibitory activities of the compounds of the present invention were evaluated in the following manner.

The antipapain activity was determined as follows. A compound of the present invention at various concentrations, 0.015 U of papain and 0.88 mg of EGTA were dissolved in 1 ml of a citrate buffer solution (20 mM, pH = 6.2). The solution so formed was preincubated at 30°C for 5 minutes. Then 1 ml of a substrate solution was added to initiate the reaction. As a substrate, a 1% solution of casein in a citrate buffer solution was employed. This reaction was conducted at 30°C for 20 minutes. Next, 3 ml of 6.5% trichloroacetic acid was added to the reaction mixture to thereby stop the reaction. The amount of proteins in the trichloroacetic acid-soluble fraction of the casein digested with the enzyme was determined by Lowry-Folin method. The obtained data were compared with control data so as to determine the inhibitory activity.

The anticalpain activity, i.e., the anticalpain I or anticalpain II activity was determined as follows. A compound of the present invention at various concentrations, 0.33 U of calpain I or II and 0.22 mg of calcium chloride were dissolved in 1 ml of an imidazole/hydrochloride buffer solution (50 mM, pH = 7.5). The solution so formed was preincubated at 30°C for 5 minutes. Then 1 ml of a substrate solution was added to initiate the reaction. As a substrate, a 0.4% solution of casein in an imidazole/hydrochloride buffer solution was employed. This reaction was conducted at 30°C for 30 minutes. Next, 3 ml of 5% trichloroacetic acid was added to stop the reaction. The amount of proteins in the trichloroacetic acid-soluble fraction of the casein digested with the enzyme was determined by Ross-Scahtz method. Th obtained data were compared with control data so as to determine the inhibitory activity.

The anticathepsin activity was determined as follows. A solution comprising a compound of the present invention at various concentrations and 0.114 mg of a substrate (benzyloxycarbonyl-L-lysine p-nitrophenyl ester) in 3.15 ml of an acetate buffer solution (25 mM, pH = 5.1, containing 1 mM of EDTA) was preincubated at 30°C for 1 minute. Then a solution of 0.05 U of cathepsin B originating from bovine spleen (Sigma Co.) dissolved in the same buffer (0.05 ml) was added to initiate the reaction. A change in th absorbance at 328 nm was monitored immediately after the initiation of the reaction. The enzyme inhibitory activity was determined by comparing the data with those obtained by use of a control solution.

Tables 2, 3, 4 and 5 show the inhibitory activities of the compounds of the. present invention respectively on papain, calpains and cathepsin B, which is the target enzyme, thus determined. In the cases of papain and calpains, calpeptin (Thujinaka et al.1 Biochem. Biophys. Res. Commun., 153, 1201 - 1208, 1988) was employed as a control.

Table 2

Inhibitory Activity on Papain	
Example No.	Inhibitory activity IC_{50} (M)
3	1.2×10^{-7}
5	2.1×10^{-7}
6	5.8×10^{-8}
7	2.5×10^{-7}
8	3.1×10^{-8}
9	1.8×10^{-7}
10	2.8×10^{-7}
12	1.8×10^{-7}
14	1.7×10^{-7}
16	9.8×10^{-8}
Calpeptin	3.1×10^{-7}

Table 3

Inhibitory Activity on Calpain I	
Example No.	Inhibitory activity IC_{50} (M)
1	1.8×10^{-7}
3	1.0×10^{-7}
4	3.0×10^{-8}
5	1.5×10^{-7}
6	8.7×10^{-8}
7	1.0×10^{-7}
8	9.2×10^{-8}
9	1.1×10^{-7}
10	9.6×10^{-8}
12	1.1×10^{-7}
14	9.0×10^{-8}
16	4.0×10^{-7}
18	7.0×10^{-7}
Calpeptin	7.1×10^{-7}

Table 4

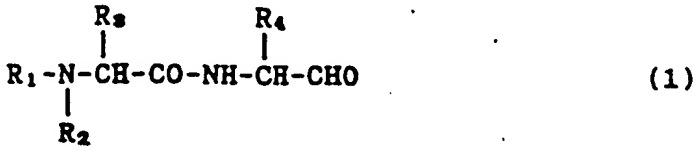
Inhibitory Activity on Calpain II	
Example No.	Inhibitory activity IC_{50} (M)
3	1.3×10^{-7}
4	1.2×10^{-7}
6	1.3×10^{-7}
7	1.3×10^{-7}
8	1.3×10^{-7}
9	1.3×10^{-7}
14	1.2×10^{-7}
Calpeptin	1.3×10^{-7}

Table 5

Inhibitory Activity on Cathepsin B	
Example No.	Inhibitory activity IC_{50} (M)
1	1.1×10^{-7}
2	9.2×10^{-6}
3	5.1×10^{-7}
4	5.8×10^{-6}
5	1.8×10^{-7}
6	7.0×10^{-6}
7	1.5×10^{-7}
8	9.9×10^{-6}
9	1.4×10^{-7}
10	3.3×10^{-7}
11	9.6×10^{-7}
12	1.2×10^{-6}
14	7.9×10^{-6}
15	3.2×10^{-6}
16	7.0×10^{-6}
17	1.0×10^{-6}
18	1.1×10^{-7}

Claims

1. A compound represented by the following general formula (1):



(1)

wherein R_1 represents a straight-chain or branched acyl group having 2 to 10 carbon atoms, a branched-, cyclic- or polycyclic-alkyloxycarbonyl group having 4 to 15 carbon atoms, a substituted- or unsubstituted-benzyloxycarbonyl group, a 2,2,2-trichloroethyloxycarbonyl group, a 2-(trimethylsilyl)ethyloxycarbonyl group, a p-toluenesulfonyl group, an o-nitrophenylsulfenyl group, a diphenylphosphonothioyl group, a triphenylmethyl group or a 2-benzoyl-1-methylvinyl group;

R_2 represents a hydrogen atom; or R_1 and R_2 may together form a phthaloyl group;

R_3 represents an isobutyl group, a n-butyl group or an isopropyl group and the above-mentioned R_1 can be an unsubstituted-benzyloxycarbonyl group provided that R_3 is a n-butyl group; and

R_4 represents a n-butyl group.

2. A compound as claimed in Claim 1, wherein R_1 is a straight-chain or branched acyl group having 2 to 10 carbon atoms selected from among octanoyl, caproyl and isovaleryl groups.

3. A compound as claimed in Claim 1, wherein R_1 is a branched-, cyclic- or polycyclic-alkyloxycarbonyl group having 4 to 15 carbon atoms selected from among t-butyloxycarbonyl, adamantyloxycarbonyl and isobornyloxycarbonyl groups.

4. A compound as claimed in Claim 1, wherein R_1 is a substituted-benzyloxycarbonyl group having one or more substituents selected from among halogen atoms, nitro group and methoxy group.

5. A proteinase inhibitor which contains a compound represented by the general formula (1) shown in Claim 1 as an active ingredient.

6. A pharmaceutical composition, comprising a compound as defined in claim 1 as an active ingredient and one or more pharmaceutically acceptable carriers.

7. Use of a compound as defined in claim 1 for making a medicament, effective in inhibiting proteinases.

European Patent Office

EUROPEAN SEARCH REPORT

Application Number

EP 90 10 6738

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.5)
Y	FEBS LETTERS, vol. 195, nos. 1,2, January 1986, pages 265-268, Federation of European Biochemical Societies, Amsterdam, NL; G. CS.-SZABO et al.: "Specific inhibition of human granulocyte elastase with peptide aldhydes" * Whole document, especially page 267, paragraph 3.2, lines 1-3 *	1-7	C 07 K 5/06 A 61 K 37/64
Y	FR-A-2 490 632 (NIPPON KAYAKU K.K.) * Whole document, in particular page 1, lines 1-12 and examples 1-37 on pages 2-4 *	1-7	
A	BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, vol. 153, no. 3, 30th June 1988, pages 1201-1208, Academic Press, Inc., San Diego, US; T. TSUJINAKA et al.: "Synthesis of a new cell penetrating calpain inhibitor (calpeptin) * Whole document *	1-7	
A	CHEMICAL ABSTRACTS, vol. 105, no. 25, 22nd December 1986, page 865, abstract no. 227326w, Columbus, Ohio, US; & JP-A-61 103 897 (SUNTORY, LTD et al.) 22-05-1986 * Abstract *	1-7	
A	DE-A-1 958 383 (ZAIDAN HOJIN) * Whole document, especially page 1, lines 1-4 *	1-7	

-/-

TECHNICAL FIELDS SEARCHED (Int. Cl.5)

C 07 K
A 61 K

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
THE HAGUE	20-06-1990	MASTURZO P.

EPO FORM 1503 03.82 (P0401)

. CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
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T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

& : member of the same patent family, corresponding document

European Patent Office

EUROPEAN SEARCH REPORT

Application Number

EP 90 10 6738

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.5)
4 A	CHEMICAL ABSTRACTS, vol. 103, no. 17, 28th October 1985, page 144, abstract no. 135650c, Columbus, Ohio, US; L. STEARDO et al.: "CCK26-33 degrading activity in brain and nonneural tissue: a metalloendopeptidase", & J. NEUROCHEM. 1985, 45(3), 784-90 * Abstract *	1-7	
			TECHNICAL FIELDS SEARCHED (Int. Cl.5)

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
THE HAGUE	20-06-1990	MASTURZO P.

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
P : intermediate document

T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

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